Remarks

Applicant encloses a check to cover the fee for additional claims in excess of twenty. Applicant believes no other fee is required with this paper; however, in the event the Examiner determines an additional fee is required, the Commissioner is authorized to charge any requisite fee or credit any overpayment to Deposit Account 20-1507. In the event a Petition for an extension of time is needed, this paper is to be considered such a Petition. Claims 2-9, and 11-40 are pending. By the Office Action mailed on January 22, 2003, by Examiner Speigler of Art Unit 1637, claims 2-9, and 11-37 stand rejected. Applicant has amended claim 37, and added new claims 39 and 40. Basis for the amendment is found throughout the specification, for example, Example 1, and pages 4-8. No new matter has been added.

I. Rejection of claims 2-9 and 11-37 under 35 U.S.C. § 112, second paragraph.

Claims 2-9 and 11-37 are rejected under 35 U.S.C. § 112, second paragraph, as allegedly failing to particularly point out and distinctly claim the subject matter which Applicant regards as the invention because various terms in independent claim 37 lack antecedent basis. The 112 rejections are presented in paragraph 3, A-D of the Action. Applicant has amended claim 37 to address each of respective rejections as follows.

A. Applicant has amended claim 37 by deleting "the other type of nucleotide". Because the allegedly ambiguous language has been deleted, the rejection is moot.

B. Applicant has amended claim 37 by inserting --extension product-- after "labeled nucleotides incorporated into a primer". This amendment clarifies that the primer extension products incorporate labeled nucleotides. In light of this amendment, Applicant submits that the rejection is overcome.

C. Applicant has amended claim 37 by deleting "high stringency conditions". Because the allegedly ambiguous language has been deleted, the rejection is moot.

D. Applicant has amended claim 37 by inserting --base-- after "nucleotide" as suggested in the Action. Thus, Applicant submits that the rejection is overcome.

II. Rejection of Claim 2-9, 11-15, and 23-37 under 35 U.S.C. § 102(b) to Applied Genetics (WO 96/30545).

Claims 2-9, 11-15, and 23-37 are rejected under 35 U.S.C. § 102(b) as anticipated because Applied Genetics allegedly discloses a method of detecting a target nucleic acid by performing a primer extension reaction in the presence of three non-terminators. The Action further alleges that mutation can be any variant, the terminator is a dideoxynucleotide, and the

non-terminators and terminators may be labeled. Applicant respectfully traverses this rejection for the reasons below.

A. Independent Claim 37.

Applicant has amended claim 37 to be directed to detecting or quantifying the presence of a nucleic acid containing a mutation of a known wild-type nucleotide by selectively forming a labeled primer extension product to detect or quantify the presence of a target nucleotide corresponding to the mutation. As discussed above, Applicant has also amended the claim to address 35 U.S.C. § 112, first paragraph concerns.

In particular, Applicant amended paragraph (e) of claim 37 to clarify that nucleotides in the reaction mixture are not complementary to the wild-type target nucleotide base. Applicant also clarified paragraph (f) of claim 37 by inserting "wherein a labeled primer extension product does not form when the target nucleotide base is wild-type". Applicant further amended the claim by inserting "wherein detecting the labeled primer extension product is not based on size". Basis for these amendments is found throughout the specification, for example, in the Examples and pages 4-9.

B. The Disclosure of Applied Genetics.

Applied Genetics discloses using primer extension reactions to detect mutations using selected mixtures of up to three nucleoside triphosphates and one or more chain terminating base pairing entities. Page 12, lines 30-35 of the reference concisely articulate the methods disclosed in the patent application as a primer extension method that "generates extension products of different lengths from related polynucleotide templates, using a primer, a polymerase enzyme, and appropriate combinations of dNTPs and, preferably, ddNTPs." The reference continues describing the methods as resolving the primer extension products based on their molecular weights or quantification by measurement of a label. The quantification of a label is performed after the primer extension products have been separated based on size. See pages 7, lines 15 and 35-37; and 20, line 35 through page 21, line 14. Page 23, lines 12-20 disclose using different fluorophors to detect different length primer extension products. Page 27, lines 23-25 provide that "the presence of the mutation is easily deduced based on the sizes of the product relative to the primer." Thus, considering the reference in its entirety, the reference discloses methods of detecting mutations by generating extension products of different sizes, and using the differences in size to detect the extension products. Applied Genetics does not disclose selecting the primer

extension reaction mixture to include (i) three types of non-terminator nucleotides that are not complementarily matched to the wild-type target nucleotide, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally (ii) one type of terminator nucleotide that is complementarily matched to the wild-type target nucleotide, wherein the terminator nucleotide is not labeled. When the primer extension reaction mixture as presently claimed is used, a labeled primer extension product does not form when the target nucleotide base is wild-type. A *labeled* primer extension product only forms if the target nucleotide base is a different base than wild-type, i.e. a mutation. Thus, the detection of a labeled primer extension product indicates that the target nucleic acid contains a mutation. Moreover, the presently claimed subject matter, unlike Applied Genetics, does not use size including length or weight to detect or quantify the primer extension products.

C. Applied Genetics Does Not Disclose All of the Elements of the Claims.

To anticipate a claim, a prior art reference must disclose every limitation of the claimed subject matter, either explicitly or inherently." In re Schreiber, 128 F.3d 1473, 1477 (Fed. Cir. 1997). Applied Genetics does not disclose all of the elements of the claims either expressly or inherently because it does not disclose selecting the primer extension reaction mixture to include (i) three types of non-terminator nucleotides that are not complementarily matched to the wild-type target nucleotide, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally (ii) one type of terminator nucleotide that is complementarily matched to the wild-type target nucleotide, wherein the terminator nucleotide is not labeled. As discussed above, when using such a primer extension reaction mixture as presently claimed, a labeled primer extension product does not form when the target nucleotide base is wild-type. A labeled primer extension product does not form when the target nucleotide is wild-type because the nucleotide base complementary to the wild-type nucleotide is purposefully omitted or is intentionally unlabeled.

Applied Genetics discloses forming primer extension products of both mutant and wild-type nucleic acids. The presence of a mutation is detected by comparing the size of the primer extension products formed. For example, page 24, lines 26-30 disclose that "the extension products derived from the wild-type templates are one base longer that those derived from the mutant templates." Pending claim 37 is directed a method that excludes detecting the labeled primer extension product based on size. Because Applied Genetics does not disclose a method

wherein a labeled primer extension product does not form when the target nucleotide base is wild-type and wherein detecting the labeled primer extension product is not based on the size of the labeled extension product, Applied Genetics does not disclose all the elements of the claim and cannot anticipate the claim. Additionally, because claims 2-9, 11-15, and 23-36 ultimately depend on claim 37, claims 2-9, 11-15 and 23-36 incorporate the limitations of claim 37 and are similarly not anticipated by Applied Genetics.

III. Rejection of Claim 2-9 and 11-37 under 35 U.S.C. § 102(e) to Soderlund (U.S. Pat. No. 6,013,431).

Claims 2-9 and 11-37 are rejected as anticipated by Soderlund because Soderlund allegedly discloses a method of detecting a target nucleic acid including (a) providing a detectable amount of target nucleic acid polymer in a single stranded form, (b) hybridizing the detectable amount of the nucleic acid polymer with one or more oligonucleotide primers (forming a primer-nucleic acid duplex), wherein each primer has a nucleotide sequence that is complementary to a sequence in the target nucleic acid polymer, such that when the primer is hybridized to the target nucleic acid polymer, the 3' end of the primer binds to a nucleotide flanking the specific nucleotide at the defined site in the target nucleic acid (i.e., the first unpaired base immediately downstream of the 3' end of the primer), (c) exposing the hybridized nucleic acid polymer to a polymerization agent in a mixture containing at least one deoxynucleotides, said deoxynucleotides comprising a detectable label, and one or more chain terminating nucleotide analogues, such that a detectable primer extension product is formed if the labeled deoxynucleotides is complementary to the specific nucleotide at the defined sites, and (d) analyzing the polymerization mixture of step (c) for the presence or absence of the primer extension product containing the labeled deoxynucleotides at the 3' end thereof, whereby the identity of the specific nucleotide at the defined site is determined.

The Action further alleges that Soderlund discloses: adding two or more differently labeled non-terminator nucleotides to the primer-nucleic acid duplex, wherein the detection is better interpreted by adding dNTPs that are different than the terminator nucleotide; using various labels such as radioactive or fluorescent labels; performing primer extension reaction by enzymatic means using template dependent enzymes; that the primer may contain an attachment moiety; that a solid support may be used in the separation process of incorporated reagent from the nucleic acid of interest; and that the nucleic acid of interest can be any human, animal, plant, or microbe. Applicant respectfully traverses this rejection.

A. The Disclosure of Soderlund.

Soderland discloses a method of sequencing a nucleic acid to determine the identity of a nucleotide base at a specific position in the nucleic acid. Generally, the method includes amplifying the target nucleic acid, annealing a primer immediately adjacent to the nucleotide base of interest, and performing primer extension by extending the primer with the nucleoside triphosphate complementary to the variable nucleotide adjacent to the primer. In essence, Soderland discloses a method of nucleotide sequencing. A labeled primer extension product is formed "if the labeled deoxynucleotide is complementary to the specific nucleotide at the defined site." Depending on which nucleotide is labeled in the primer extension reaction mixture, the detection of the label in the primer extension product indicates the identity of the nucleotide base of interest.

For example, Soderlund discloses using one labeled terminator nucleotide complementary to X1 or X2. The primer extension product terminates after the incorporation of the labeled terminator nucleotide. Soderlund explains further that this embodiment is suitable for use by dividing a sample into two parts and detecting for X1 in one part and X2 in another part. Because the labeled terminator nucleotide is known, the identity of variable nucleotide is known depending on which part of the sample produced a labeled primer extension product.

Soderlund also discloses that two differently labeled terminator nucleotides can be used. Because the identity of the nucleotide with each label is known, the variable nucleotide in a primer extension product incorporating one of the labeled terminator nucleotides can be identified.

Soderlund further discloses that one labeled deoxynucleotide can be used optionally with a terminator nucleotide, or two or more types of differently labeled nucleotides can be used. Thus, the incorporation of the labeled deoxynucleotide into part of a sample can determine the identity of the variable nucleotide in that sample if the variable nucleotide is complementary to the labeled deoxynucleotides.

The central concept through all of Soderlund's embodiments is correlating the identity of a labeled nucleotide incorporated into a primer extension product with the identity of a variable nucleotide. In every embodiment, it is critical to add a labeled nucleotide corresponding to either X1 or X2 to form a detectable primer extension product. Soderlund does not disclose a primer reaction method in which the primer reaction mixture is formed by selecting three types

of non-terminator nucleotides that are not complementarily matched to a wild-type target nucleotide, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally selecting one type of non-labeled terminator nucleotide that is complementarily matched to the wild-type target nucleotide such that a labeled primer extension product does not form when the target nucleotide base is a wild-type nucleotide base. In sum, Soderlund does not disclose omitting a nucleotide base, labeled or unlabeled, that is complementary to a known wild-type nucleotide thereby selectively forming a labeled primer extension product corresponding to a mutation of the known wild-type nucleotide.

B. Soderlund does not disclose all of the elements of the claims.

Independent claim 37 is described above and contains elements that are not disclosed in Soderlund. The Office Action suggests that the present patent application is directed to determining the identity of an unknown base at a predetermine location. Applicant respectfully disagrees. Claim 37 and dependent claims 2-9 and 11-36 are directed to a method for detecting or quantifying the presence of a target nucleic acid containing a mutation of a *known* wild-type nucleotide in a sample by detecting a signal from a plurality of labeled nucleotides incorporated into a primer extension product. Soderlund discloses forming labeled primer extension products to determine the identity of an *unknown* variable nucleotide.

Soderlund does not disclose, for example, mixing the primer-nucleic acid duplex with a primer extension reaction reagent comprising: (i) three types of non-terminator nucleotides that are not complementarily matched to the *known* wild-type target nucleotide, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally (ii) one type of terminator nucleotide that is complementarily matched to the known wild-type target nucleotide, wherein the terminator nucleotide is not labeled. Additionally, Soderlund does not disclose extending the primer extension reaction by enzymatic or chemical means to form a labeled primer extension product comprising a plurality of labeled non-terminator nucleotides, wherein a labeled primer extension product does not form when the target nucleotide base is wild-type.

Using the reaction mixture presently claimed results in the selective formation of a labeled primer extension product when the target nucleic acid contains a mutated target nucleotide base of a known wild-type nucleotide. In the case where the nucleotide base complementary to the wild-type target nucleotide base is omitted, a labeled primer extension will

not form when the nucleic acid contains a wild-type target nucleotide base at the specified position because the wild-type nucleotide base is the first unpaired base immediately downstream of the 3' end of the primer. In the case when an unlabeled terminator nucleotide is complementary to the wild-type nucleotide, an unlabeled, single nucleotide extension product will be formed when the target nucleic acid contains a wild-type target nucleotide base, but a labeled extension product will not be formed because the wild-type nucleotide is the first unpaired base immediately downstream of the 3' end of the primer. Thus, in this last case, the unlabeled terminator nucleotide prevents the formation of a labeled primer extension product. Because Soderlund does not disclose all of the elements of the claims, the reference cannot anticipate the claims.

IV. Rejection of Claims 16-22 under 35 U.S.C. § 103(a) over Applied Genetics in view of Shuber (U.S. Pat. No. 5,888,778).

Claims 16-22 are rejected as obvious over Applied Genetics in view of Shuber because it would allegedly be obvious to one of ordinary skill in the art at the time the invention was made to have modified the method of Applied Genetics so as to have immobilized a primer on a solid-phase, instead of carrying out the method in solution, in order to have achieved the benefits of using solid-phase supports as taught by Shuber of simultaneously processing and screening a large number of samples and controls, and thus facilitating analysis, and furthermore, that solid-phase supports can be used in automated systems. Applicant respectfully traverses this rejection.

A. The Teachings of Shuber

Shuber generally teaches methods for high-throughput screening to detect genetic alterations in a sample. Nucleic acids to be screened are attached to a solid support, for example a microtiter dish. Single-base extension is performed on the nucleic acid with a segmented primer. The extension products are isolated and sequenced. Shuber does not teach or suggest detecting or quantifying a nucleic acid containing a mutation of a known wild-type nucleotide by selectively forming a labeled primer extension product to detect or quantify the presence of a target nucleotide corresponding to the mutation. Additionally, Shuber does not teach or suggest a primer reaction method in which the primer reaction mixture is formed by selecting three types of non-terminator nucleotides that are not complementarily matched to a wild-type target nucleotide, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally selecting one type of non-labeled terminator nucleotide that is

complementarily matched to the wild-type target nucleotide such that a labeled primer extension product does not form when the target nucleotide base is wild-type nucleotide base.

B. Applied Genetics Does Not Teach or Suggest All of the Elements of the Claimed Subject Matter.

Because claims 16-22 ultimately depend on claim 37, claims 16-22 incorporate the elements of claim 37. As discussed above in view of claim 37, Applied Genetics does not disclose all of the elements of the presently claimed subject matter. Moreover, Applied Genetics does not teach or suggest all of the elements of the claimed subject matter. In particular, Applied Genetics does not teach or suggest a method wherein a labeled primer extension product does not form when the target nucleotide base is wild-type and wherein detecting the labeled primer extension product is not based on the size of the labeled extension product. More specifically, Applied Genetics does not teach or suggest a primer extension reaction mixture including (i) three types of non-terminator nucleotides that are not complementarily matched to a specific wild-type target nucleotide, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally (ii) one type of terminator nucleotide that is complementarily matched to the wild-type target nucleotide, wherein the terminator nucleotide is not labeled. Because Applied Genetics does not teach or suggest all of the elements of the claimed subject matter, it cannot render the claimed subject matter obvious.

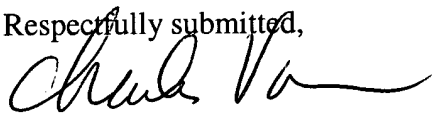
C. Shuber Does Not Cure the Deficiencies of Applied Genetics.

As discussed above, Shuber teaches methods for high-throughput screening to detect genetic alterations in a sample. Shuber does not teach or suggest a primer extension reaction mixture including (i) three types of non-terminator nucleotides that are not complementarily matched to a specific wild-type target nucleotide, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally (ii) one type of terminator nucleotide that is complementarily matched to the wild-type target nucleotide, wherein the terminator nucleotide is not labeled. Thus, Shuber does not cure the deficiencies of Applied Genetics, and the combination of Applied Genetics and Shuber cannot render the claims obvious.

Conclusions

In light of the forgoing Amendments and Remarks, Applicant respectfully requests reconsideration and allowance of all pending claims. The undersigned invites the Examiner to call 404-885-3813 to further discuss this response should the Examiner have any additional concerns.

Respectfully submitted,



Charles Vorndran
Reg. No. 45,315

Troutman Sanders LLP
600 Peachtree Street, N.E.
Suite 5200
Atlanta, Georgia 30308
404-885-3813
404-962-6866 fax